



# SEQUENCE LISTING

<110> ISHIKAWA, KOHKI  
SUZUKI, EI-ICHIRO  
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SHIMBA, NOBUHISA  
MIHARA, YASUHIRO  
KAWASAKI, HISASHI  
KURAHASHI, OSAMU  
KOURA, TOHRU  
SHIMAOKA, MEGUMI  
KOZUTSUMI, RIE  
ASANO, YASUHISA

<120> MUTANT NUCLEOSIDE-5'-PHOSPHATE PRODUCING ENZYMES

<130> 206523US0PCT

<140> 09/807,990

<141> 2000-09-01

<150> JP 11/249545

<151> 1999-09-03

<160> 125

<170> PatentIn version 3.3

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cgcggggggtt ccccggggccc ctttttttta tggggctgcg gtgaggagcg ttatctgctg 180  
gccctgtttg tgcaacaaac gctttttattg tgtaattttt gtgacgtata tcagggtttt 240  
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Met Lys Lys Arg Val Leu Ala Val

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5

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| tgt ttt gcc gca ttg ttc tct tct cag gcc ctg gcg ctg gtc gct acc | 402 |
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| ggc aac gac act acc acg aaa ccg gat ctc tac tac ctc aag aac agt | 450 |
| Gly Asn Asp Thr Thr Thr Lys Pro Asp Leu Tyr Tyr Leu Lys Asn Ser |     |
| 25 30 35 40   |     |
| gaa gcc att aac agc ctg gcg ctg ttg ccg cca cca ccg gcg gtg ggc | 498 |
| Glu Ala Ile Asn Ser Leu Ala Leu Leu Pro Pro Pro Pro Ala Val Gly |     |
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| tcc att gcg ttt ctc aac gat cag gcc atg tat gaa cag ggg cgc ctg | 546 |
| Ser Ile Ala Phe Leu Asn Asp Gln Ala Met Tyr Glu Gln Gly Arg Leu |     |
| 60 65 70  |     |
| ctg cgc aac acc gaa cgc ggt aag ctg gcg gcg gaa gat gca aac ctg | 594 |
| Leu Arg Asn Thr Glu Arg Gly Lys Leu Ala Ala Glu Asp Ala Asn Leu |     |
| 75 80 85  |     |
| agc agt ggc ggg gtg gcg aat gct ttc tcc ggc gcg ttt ggt agc ccg | 642 |
| Ser Ser Gly Gly Val Ala Asn Ala Phe Ser Gly Ala Phe Gly Ser Pro |     |
| 90 95 100   |     |
| atc acc gaa aaa gac gcc ccg gcg ctg cat aaa tta ctg acc aat atg | 690 |
| Ile Thr Glu Lys Asp Ala Pro Ala Leu His Lys Leu Leu Thr Asn Met |     |
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| att gag gac gcc ggg gat ctg gcg acc cgc agc gcg aaa gat cac tat | 738 |
| Ile Glu Asp Ala Gly Asp Leu Ala Thr Arg Ser Ala Lys Asp His Tyr |     |
| 125 130 135   |     |
| atg cgc att cgt ccg ttc gcg ttt tat ggg gtc tct acc tgt aat acc | 786 |
| Met Arg Ile Arg Pro Phe Ala Phe Tyr Gly Val Ser Thr Cys Asn Thr |     |
| 140 145 150   |     |
| acc gag cag gac aaa ctg tcc aaa aat ggc tct tat ccg tcc ggg cat | 834 |
| Thr Glu Gln Asp Lys Leu Ser Lys Asn Gly Ser Tyr Pro Ser Gly His |     |
| 155 160 165   |     |
| acc tct atc ggc tgg gct act gcg ctg gtg ctg gca gag atc aac cct | 882 |
| Thr Ser Ile Gly Trp Ala Thr Ala Leu Val Leu Ala Glu Ile Asn Pro |     |
| 170 175 180   |     |
| cag cgc cag aac gag atc ctg aaa cgc ggt tat gag ctg ggc cag agc | 930 |
| Gln Arg Gln Asn Glu Ile Leu Lys Arg Gly Tyr Glu Leu Gly Gln Ser |     |
| 185 190 195 200   |     |
| cgg gtg att tgc ggc tac cac tgg cag agt gat gtg gat gcc gcg ccg | 978 |

Arg Val Ile Cys Gly Tyr His Trp Gln Ser Asp Val Asp Ala Ala Arg  
 205 210 215

gta gtg gga tct gcc gtt gtg gcg acc ctg cat acc aac ccg gcg ttc 1026  
 Val Val Gly Ser Ala Val Val Ala Thr Leu His Thr Asn Pro Ala Phe  
 220 225 230

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 235 240 245

aaa taatcctgac taccgccttg ccttgcaggg cggtagtggg ttccactggc 1127  
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Leu Pro Pro Pro Pro Ala Val Gly Ser Ile Ala Phe Leu Asn Asp Gln  
 50 55 60

Ala Met Tyr Glu Gln Gly Arg Leu Leu Arg Asn Thr Glu Arg Gly Lys  
 65 70 75 80

Leu Ala Ala Glu Asp Ala Asn Leu Ser Ser Gly Gly Val Ala Asn Ala  
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Phe Ser Gly Ala Phe Gly Ser Pro Ile Thr Glu Lys Asp Ala Pro Ala  
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Leu His Lys Leu Leu Thr Asn Met Ile Glu Asp Ala Gly Asp Leu Ala  
 115 120 125

Thr Arg Ser Ala Lys Asp His Tyr Met Arg Ile Arg Pro Phe Ala Phe  
 130 135 140

Tyr Gly Val Ser Thr Cys Asn Thr Thr Glu Gln Asp Lys Leu Ser Lys  
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Asn Gly Ser Tyr Pro Ser Gly His Thr Ser Ile Gly Trp Ala Thr Ala  
 165 170 175

Leu Val Leu Ala Glu Ile Asn Pro Gln Arg Gln Asn Glu Ile Leu Lys  
 180 185 190

Arg Gly Tyr Glu Leu Gly Gln Ser Arg Val Ile Cys Gly Tyr His Trp  
 195 200 205

Gln Ser Asp Val Asp Ala Ala Arg Val Val Gly Ser Ala Val Val Ala  
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Ala Glu Phe Ala Gln His Gln Lys Lys  
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| cttattttatc cgttcggttaa caaaagccat gctgtttctg tcaaattatc tgaaaatcat | 180 |
| catcaaaaat acttacctgt cttccgtctg tttcgtcaca cttttttgaa agagttaaca   | 240 |
| tcaatttgca tctctccgcc ctacactggc agacagggtt ctgagtaata ctgttgatc    | 300 |
| tgataaggag atgtc atg aag aag aat att atc gcc ggt tgt ctg ttc tca    | 351 |
| Met Lys Lys Asn Ile Ile Ala Gly Cys Leu Phe Ser                     |     |
| 1 5 10  |     |
| ctg ttt tcc ctt tcc gcg ctg gcc gcg atc ccg gcg ggc aac gat gcc     | 399 |
| Leu Phe Ser Leu Ser Ala Leu Ala Ala Ile Pro Ala Gly Asn Asp Ala     |     |
| 15 20 25  |     |
| acc acc aag ccg gat tta tat tat ctg aaa aat gaa cag gct atc gac     | 447 |
| Thr Thr Lys Pro Asp Leu Tyr Tyr Leu Lys Asn Glu Gln Ala Ile Asp     |     |
| 30 35 40  |     |
| agc ctg aaa ctg tta ccg cca ccg ccg gaa gtc ggc agt att cag ttt     | 495 |
| Ser Leu Lys Leu Leu Pro Pro Pro Pro Glu Val Gly Ser Ile Gln Phe     |     |
| 45 50 55 60   |     |
| tta aat gat cag gca atg tat gag aaa ggc cgt atg ctg cgc aat acc     | 543 |
| Leu Asn Asp Gln Ala Met Tyr Glu Lys Gly Arg Met Leu Arg Asn Thr     |     |
| 65 70 75  |     |
| gag cgc gga aaa cag gca cag gca gat gct gac ctg gcc gca ggg ggt     | 591 |
| Glu Arg Gly Lys Gln Ala Gln Ala Asp Ala Asp Leu Ala Ala Gly Gly     |     |
| 80 85 90  |     |
| gtg gca acc gca ttt tca ggg gca ttc ggc tat ccg ata acc gaa aaa     | 639 |
| Val Ala Thr Ala Phe Ser Gly Ala Phe Gly Tyr Pro Ile Thr Glu Lys     |     |
| 95 100 105  |     |
| gac tct ccg gag ctg tat aaa ctg ctg acc aat atg att gag gat gcc     | 687 |
| Asp Ser Pro Glu Leu Tyr Lys Leu Leu Thr Asn Met Ile Glu Asp Ala     |     |
| 110 115 120   |     |
| ggt gat ctt gcc acc cgc tcc gcc aaa gaa cat tac atg cgc atc cgg     | 735 |
| Gly Asp Leu Ala Thr Arg Ser Ala Lys Glu His Tyr Met Arg Ile Arg     |     |
| 125 130 135 140   |     |
| ccg ttt gcg ttt tac ggc aca gaa acc tgt aat acc aaa gat cag aaa     | 783 |
| Pro Phe Ala Phe Tyr Gly Thr Glu Thr Cys Asn Thr Lys Asp Gln Lys     |     |
| 145 150 155   |     |
| aaa ctc tcc acc aac gga tct tac ccg tca ggt cat acg tct atc ggc     | 831 |

Lys Leu Ser Thr Asn Gly Ser Tyr Pro Ser Gly His Thr Ser Ile Gly  
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 Ala Ile Leu Glu Arg Gly Tyr Gln Leu Gly Gln Ser Arg Val Ile Cys  
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 Gly Tyr His Trp Gln Ser Asp Val Asp Ala Ala Arg Ile Val Gly Ser  
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 Ala Ala Val Ala Thr Leu His Ser Asp Pro Ala Phe Gln Ala Gln Leu  
 225 230 235  
 gcg aaa gcc aaa cag gaa ttt gca caa aaa tca cag aaa taaaagcagt 1072  
 Ala Lys Ala Lys Gln Glu Phe Ala Gln Lys Ser Gln Lys  
 240 245  
 gatattctggg cagggcagtg caatatctgc cctgaaatcc ctgtttattc ccacatccag 1132  
 cgggtcttccc gatcccagcc ttttgttttc atgcagctgt agaaatagcg gttgcggctg 1192  
 tcttcattca catccatcac ataactttcc gttaccggtg tctgctcttt gtaggttttg 1252  
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Asp Leu Tyr Tyr Leu Lys Asn Glu Gln Ala Ile Asp Ser Leu Lys Leu  
 35 40 45

Leu Pro Pro Pro Pro Glu Val Gly Ser Ile Gln Phe Leu Asn Asp Gln  
 50 55 60

Ala Met Tyr Glu Lys Gly Arg Met Leu Arg Asn Thr Glu Arg Gly Lys  
 65 70 75 80

Gln Ala Gln Ala Asp Ala Asp Leu Ala Ala Gly Gly Val Ala Thr Ala  
 85 90 95

Phe Ser Gly Ala Phe Gly Tyr Pro Ile Thr Glu Lys Asp Ser Pro Glu  
 100 105 110

Leu Tyr Lys Leu Leu Thr Asn Met Ile Glu Asp Ala Gly Asp Leu Ala  
 115 120 125

Thr Arg Ser Ala Lys Glu His Tyr Met Arg Ile Arg Pro Phe Ala Phe  
 130 135 140

Tyr Gly Thr Glu Thr Cys Asn Thr Lys Asp Gln Lys Lys Leu Ser Thr  
 145 150 155 160

Asn Gly Ser Tyr Pro Ser Gly His Thr Ser Ile Gly Trp Ala Thr Ala  
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Leu Val Leu Ala Glu Val Asn Pro Ala Asn Gln Asp Ala Ile Leu Glu  
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Arg Gly Tyr Gln Leu Gly Gln Ser Arg Val Ile Cys Gly Tyr His Trp  
 195 200 205

Gln Ser Asp Val Asp Ala Ala Arg Ile Val Gly Ser Ala Ala Val Ala  
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Gln Glu Phe Ala Gln Lys Ser Gln Lys

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 agtgagtctt t atg aaa agt cgt tat tta gta ttt ttt cta cca ctg atc 170  
                   Met Lys Ser Arg Tyr Leu Val Phe Phe Leu Pro Leu Ile  
                   1                  5                  10  
 gta gct aaa tat aca tca gca gaa aca gtg caa ccc ttt cat tct cct 218  
 Val Ala Lys Tyr Thr Ser Ala Glu Thr Val Gln Pro Phe His Ser Pro  
                   15                  20                  25  
 gaa gaa tca gtg aac agt cag ttc tac tta cca cca ccg cca ggt aat 266  
 Glu Glu Ser Val Asn Ser Gln Phe Tyr Leu Pro Pro Pro Pro Gly Asn  
                   30                  35                  40                  45  
 gat gat ccg gct tac cgc tat gat aag gag gct tat ttt aag ggc tat 314  
 Asp Asp Pro Ala Tyr Arg Tyr Asp Lys Glu Ala Tyr Phe Lys Gly Tyr  
                   50                  55                  60  
 gcg ata aag ggt tcc ccg cga tgg aaa caa gct gct gag gat gca gat 362  
 Ala Ile Lys Gly Ser Pro Arg Trp Lys Gln Ala Ala Glu Asp Ala Asp  
                   65                  70                  75  
 gta agc gtg gaa aat ata gcc aga ata ttc tcg cca gta gtg ggt gct 410  
 Val Ser Val Glu Asn Ile Ala Arg Ile Phe Ser Pro Val Val Gly Ala  
                   80                  85                  90  
 aaa att aac ccc aaa gat acg cca gaa acc tgg aat atg tta aag aat 458  
 Lys Ile Asn Pro Lys Asp Thr Pro Glu Thr Trp Asn Met Leu Lys Asn  
                   95                  100                  105  
 ctt ctg aca atg ggc ggc tac tac gct act gct tcg gca aaa aaa tat 506  
 Leu Leu Thr Met Gly Gly Tyr Tyr Ala Thr Ala Ser Ala Lys Lys Tyr  
                   110                  115                  120                  125  
 tat atg cgt acc cgc ccc ttt gtc tta ttt aat cat tcc acc tgc cgt 554



|            |            |             |            |            |            |     |     |     |     |     |            |            |     |     |     |     |
|------------|------------|-------------|------------|------------|------------|-----|-----|-----|-----|-----|------------|------------|-----|-----|-----|-----|
| Tyr        | Met        | Arg         | Thr        | Arg        | Pro        | Phe | Val | Leu | Phe | Asn | His        | Ser        | Thr | Cys | Arg |     |
|            |            |             |            | 130        |            |     |     |     | 135 |     |            |            |     | 140 |     |     |
| cct        | gaa        | gat         | gag        | aat        | act        | ttg | cga | aaa | aat | ggc | tct        | tac        | cct | tcc | ggg | 602 |
| Pro        | Glu        | Asp         | Glu        | Asn        | Thr        | Leu | Arg | Lys | Asn | Gly | Ser        | Tyr        | Pro | Ser | Gly |     |
|            |            |             | 145        |            |            |     |     | 150 |     |     |            |            | 155 |     |     |     |
| cat        | act        | gct         | tat        | ggg        | aca        | ctt | ctg | gca | tta | gta | tta        | tcc        | gag | gcc | aga | 650 |
| His        | Thr        | Ala         | Tyr        | Gly        | Thr        | Leu | Leu | Ala | Leu | Val | Leu        | Ser        | Glu | Ala | Arg |     |
|            |            | 160         |            |            |            |     | 165 |     |     |     |            | 170        |     |     |     |     |
| ccg        | gaa        | cgc         | gcg        | cag        | gag        | ctc | gcc | aga | cgc | gga | tgg        | gag        | ttc | ggg | caa | 698 |
| Pro        | Glu        | Arg         | Ala        | Gln        | Glu        | Leu | Ala | Arg | Arg | Gly | Trp        | Glu        | Phe | Gly | Gln |     |
|            | 175        |             |            |            |            | 180 |     |     |     |     | 185        |            |     |     |     |     |
| agc        | aga        | gtg         | ata        | tgc        | ggg        | gct | cac | tgg | caa | agc | gat        | gtt        | gat | gct | ggc | 746 |
| Ser        | Arg        | Val         | Ile        | Cys        | Gly        | Ala | His | Trp | Gln | Ser | Asp        | Val        | Asp | Ala | Gly |     |
| 190        |            |             |            |            | 195        |     |     |     |     | 200 |            |            |     |     | 205 |     |
| cgt        | tat        | gtg         | gga        | gca        | gta        | gag | ttt | gca | aga | ctg | caa        | aca        | atc | ccg | gct | 794 |
| Arg        | Tyr        | Val         | Gly        | Ala        | Val        | Glu | Phe | Ala | Arg | Leu | Gln        | Thr        | Ile | Pro | Ala |     |
|            |            |             | 210        |            |            |     |     | 215 |     |     |            |            |     | 220 |     |     |
| ttt        | cag        | aag         | tca        | ctg        | gca        | aaa | tcc | gtg | agg | agc | tgaacgacaa | aaataattta |     |     |     | 847 |
| Phe        | Gln        | Lys         | Ser        | Leu        | Ala        | Lys | Ser | Val | Arg | Ser |            |            |     |     |     |     |
|            |            | 225         |            |            |            |     | 230 |     |     |     |            |            |     |     |     |     |
| ttgagtaaag | aagatcaccc | caaaacttaat | tactgaaggt | gaaagtcttc | ccgcaaactg |     |     |     |     |     |            |            |     |     |     | 907 |
| gccacagcaa | atgaaaggaa | gtgcaactgc  | gtaggggcgg | ccgggcgtgg | agaatgcctt |     |     |     |     |     |            |            |     |     |     | 967 |
| tggtttcccc | gattcgcatg | aatt        |            |            |            |     |     |     |     |     |            |            |     |     |     | 991 |

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| Met | Lys | Ser | Arg | Tyr | Leu | Val | Phe | Phe | Leu | Pro | Leu | Ile | Val | Ala | Lys |
| 1   |     |     |     | 5   |     |     |     |     | 10  |     |     |     |     | 15  |     |

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|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Thr | Ser | Ala | Glu | Thr | Val | Gln | Pro | Phe | His | Ser | Pro | Glu | Glu | Ser |
|     |     |     | 20  |     |     |     |     | 25  |     |     |     |     | 30  |     |     |

|     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Val | Asn | Ser | Gln | Phe | Tyr | Leu | Pro | Pro | Pro | Pro | Gly | Asn | Asp | Asp | Pro |
|     |     | 35  |     |     |     |     | 40  |     |     |     |     | 45  |     |     |     |

Ala Tyr Arg Tyr Asp Lys Glu Ala Tyr Phe Lys Gly Tyr Ala Ile Lys  
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Gly Ser Pro Arg Trp Lys Gln Ala Ala Glu Asp Ala Asp Val Ser Val  
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Glu Asn Ile Ala Arg Ile Phe Ser Pro Val Val Gly Ala Lys Ile Asn  
85 90 95

Pro Lys Asp Thr Pro Glu Thr Trp Asn Met Leu Lys Asn Leu Leu Thr  
100 105 110

Met Gly Gly Tyr Tyr Ala Thr Ala Ser Ala Lys Lys Tyr Tyr Met Arg  
115 120 125

Thr Arg Pro Phe Val Leu Phe Asn His Ser Thr Cys Arg Pro Glu Asp  
130 135 140

Glu Asn Thr Leu Arg Lys Asn Gly Ser Tyr Pro Ser Gly His Thr Ala  
145 150 155 160

Tyr Gly Thr Leu Leu Ala Leu Val Leu Ser Glu Ala Arg Pro Glu Arg  
165 170 175

Ala Gln Glu Leu Ala Arg Arg Gly Trp Glu Phe Gly Gln Ser Arg Val  
180 185 190

Ile Cys Gly Ala His Trp Gln Ser Asp Val Asp Ala Gly Arg Tyr Val  
195 200 205

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Ser Leu Ala Lys Ser Val Arg Ser  
225 230

<210> 7

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<220>  
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 agctatggct tccacacgat agacccgcgc aacacataat tgtcttatta tagccacatg 180  
 atatTTTTat attacaattt taaactaaaa ttaagaatta aattcttgaa ataaagggtt 240  
 ttttattaaa aggataggaa atgtcgtgaa atcggcattt tctatccata ttatataaca 300  
 aggggaagact gacgac atg ata aaa gtc ccg cgg ttc atc tgt atg atc gcg 352  
                   Met Ile Lys Val Pro Arg Phe Ile Cys Met Ile Ala  
                   1                  5                  10  
 ctt aca tcc ggc gtt ctg gca agc ggc ctt tct caa agc gtt tca gct 400  
 Leu Thr Ser Gly Val Leu Ala Ser Gly Leu Ser Gln Ser Val Ser Ala  
                   15                  20                  25  
 cat aca gaa aaa agt gaa ccc tcc tcg act tat cat ttc cac agc gat 448  
 His Thr Glu Lys Ser Glu Pro Ser Ser Thr Tyr His Phe His Ser Asp  
                   30                  35                  40  
 ccc ctt ctt tac ctt gcg ccc cca ccc act tcc ggc agt cca tta cag 496  
 Pro Leu Leu Tyr Leu Ala Pro Pro Pro Thr Ser Gly Ser Pro Leu Gln  
                   45                  50                  55                  60  
 gcg cat gat gat caa acc ttt aac agc acc aga caa tta aaa ggt agc 544  
 Ala His Asp Asp Gln Thr Phe Asn Ser Thr Arg Gln Leu Lys Gly Ser  
                   65                  70                  75  
 acg cgc tgg gca ttg gca act caa gat gcc gat ctt cat ctc gct tca 592  
 Thr Arg Trp Ala Leu Ala Thr Gln Asp Ala Asp Leu His Leu Ala Ser  
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 Val Leu Lys Asp Tyr Ala Cys Ala Ala Gly Met Asn Leu Asp Ile Ala  
                   95                  100                  105  
 caa tta ccg cat ctt gcc aat ttg att aaa cgc gca ctt cgc acc gaa 688  
 Gln Leu Pro His Leu Ala Asn Leu Ile Lys Arg Ala Leu Arg Thr Glu  
                   110                  115                  120

|   |      |
|---|------|
| tat gac gat att ggc aga gcc aaa aat aac tgg aat cgc aaa cga cct   | 736  |
| Tyr Asp Asp Ile Gly Arg Ala Lys Asn Asn Trp Asn Arg Lys Arg Pro   |      |
| 125 130 135 140   |      |
| ttt gtg gat acc gat caa ccc atc tgc acg gaa aaa gat cgc gaa ggt   | 784  |
| Phe Val Asp Thr Asp Gln Pro Ile Cys Thr Glu Lys Asp Arg Glu Gly   |      |
| 145 150 155   |      |
| ctg gga aaa caa ggc tcc tat cct tca ggt cat acg act atc ggt tgg   | 832  |
| Leu Gly Lys Gln Gly Ser Tyr Pro Ser Gly His Thr Thr Ile Gly Trp   |      |
| 160 165 170   |      |
| agc gtt gcg ctc att ctg gct gaa ttg atc ccc gat cat gcg gcg aat   | 880  |
| Ser Val Ala Leu Ile Leu Ala Glu Leu Ile Pro Asp His Ala Ala Asn   |      |
| 175 180 185   |      |
| att ttg cag cgt ggc caa att ttt gga acc agc cgg att gtc tgc ggc   | 928  |
| Ile Leu Gln Arg Gly Gln Ile Phe Gly Thr Ser Arg Ile Val Cys Gly   |      |
| 190 195 200   |      |
| gcc cat tgg ttc agc gat gtg cag gca ggc tat atc atg gca tcg ggc   | 976  |
| Ala His Trp Phe Ser Asp Val Gln Ala Gly Tyr Ile Met Ala Ser Gly   |      |
| 205 210 215 220   |      |
| gaa att gca gct tta cat ggg gat gcc gat ttc cgc cga gat atg gaa   | 1024 |
| Glu Ile Ala Ala Leu His Gly Asp Ala Asp Phe Arg Arg Asp Met Glu   |      |
| 225 230 235   |      |
| tta gct cgg aaa gaa tta gaa aag gca cgc aca tca gcg cac acg cca   | 1072 |
| Leu Ala Arg Lys Glu Leu Glu Lys Ala Arg Thr Ser Ala His Thr Pro   |      |
| 240 245 250   |      |
| gac gat ctt cta tgc aag att gaa caa agc gct cgc taaattcaat        | 1118 |
| Asp Asp Leu Leu Cys Lys Ile Glu Gln Ser Ala Arg                   |      |
| 255 260   |      |
| caagtattat ttcaacaagg ggaaagattg cttgctgtaa tttttggata tcaaacaggc | 1178 |
| gaaaaaatga aagagcgcac gctctttcaa aggcaattcg atttagtccg gtggcattct | 1238 |
| cacgccacaa accaaatcat aaataaccgc ctcttttccg ccagataact gcaaaattat | 1298 |
| agaataccga cagctggaat atcgtcactt ttcctag                          | 1335 |

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 <213> Zymomonas mobilis

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Met Ile Lys Val Pro Arg Phe Ile Cys Met Ile Ala Leu Thr Ser Gly  
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Val Leu Ala Ser Gly Leu Ser Gln Ser Val Ser Ala His Thr Glu Lys  
20 25 30

Ser Glu Pro Ser Ser Thr Tyr His Phe His Ser Asp Pro Leu Leu Tyr  
35 40 45

Leu Ala Pro Pro Pro Thr Ser Gly Ser Pro Leu Gln Ala His Asp Asp  
50 55 60

Gln Thr Phe Asn Ser Thr Arg Gln Leu Lys Gly Ser Thr Arg Trp Ala  
65 70 75 80

Leu Ala Thr Gln Asp Ala Asp Leu His Leu Ala Ser Val Leu Lys Asp  
85 90 95

Tyr Ala Cys Ala Ala Gly Met Asn Leu Asp Ile Ala Gln Leu Pro His  
100 105 110

Leu Ala Asn Leu Ile Lys Arg Ala Leu Arg Thr Glu Tyr Asp Asp Ile  
115 120 125

Gly Arg Ala Lys Asn Asn Trp Asn Arg Lys Arg Pro Phe Val Asp Thr  
130 135 140

Asp Gln Pro Ile Cys Thr Glu Lys Asp Arg Glu Gly Leu Gly Lys Gln  
145 150 155 160

Gly Ser Tyr Pro Ser Gly His Thr Thr Ile Gly Trp Ser Val Ala Leu  
165 170 175

Ile Leu Ala Glu Leu Ile Pro Asp His Ala Ala Asn Ile Leu Gln Arg  
180 185 190

Gly Gln Ile Phe Gly Thr Ser Arg Ile Val Cys Gly Ala His Trp Phe

195

200

205

Ser Asp Val Gln Ala Gly Tyr Ile Met Ala Ser Gly Glu Ile Ala Ala  
 210 215 220

Leu His Gly Asp Ala Asp Phe Arg Arg Asp Met Glu Leu Ala Arg Lys  
 225 230 235 240

Glu Leu Glu Lys Ala Arg Thr Ser Ala His Thr Pro Asp Asp Leu Leu  
 245 250 255

Cys Lys Ile Glu Gln Ser Ala Arg  
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 catctcctgt cattgcaatc ccgctatggg agcgcccaaa cggcaagggtg ataagtgcga 180  
 cagtccgaaa tcgcgagtgg ttgctcatta agcagacaaa tatgcgtttt tgcgataccg 240  
 aacaattttt tcaatgtgat tttaactttt acttacagat gacaaaaatg tgactaaaaa 300  
 caaaaccatt gttctggaca tataacaccg taaggaaatg tag atg aaa aag cgc 355  
 Met Lys Lys Arg  
 -20

|   |     |
|---|-----|
| ggt ctc gcc ctc tgc ctc gcc agc ctg ttt tcc gtt aac gct ttc gcg | 403 |
| Val Leu Ala Leu Cys Leu Ala Ser Leu Phe Ser Val Asn Ala Phe Ala |     |
| -15 -10 -5 -1   |     |
| ctg gtc cct gcc ggc aat gat gca acc acc aaa ccg gat ctc tat tat | 451 |
| Leu Val Pro Ala Gly Asn Asp Ala Thr Thr Lys Pro Asp Leu Tyr Tyr |     |
| 1 5 10 15   |     |
| ctg aaa aat gca cag gcc atc gat agt ctg gcg ctg ttg ccg ccg ccg | 499 |
| Leu Lys Asn Ala Gln Ala Ile Asp Ser Leu Ala Leu Leu Pro Pro Pro |     |
| 20 25 30  |     |
| ccg gaa gtt ggc agc atc gca ttt tta aac gat cag gcg atg tat gag | 547 |
| Pro Glu Val Gly Ser Ile Ala Phe Leu Asn Asp Gln Ala Met Tyr Glu |     |
| 35 40 45  |     |
| aaa gga cgg ctg ttg cgc aat acc gaa cgt ggc aag cag gcg cag gca | 595 |
| Lys Gly Arg Leu Leu Arg Asn Thr Glu Arg Gly Lys Gln Ala Gln Ala |     |
| 50 55 60  |     |
| gat gct gac ctg gcc gcc ggc gac gtc gcg aat gcc ttc tcc agc gct | 643 |
| Asp Ala Asp Leu Ala Ala Gly Asp Val Ala Asn Ala Phe Ser Ser Ala |     |
| 65 70 75 80   |     |
| ttt ggt tcg ccc atc acc gaa aaa gac gcg ccg cag tta cat aag ctg | 691 |
| Phe Gly Ser Pro Ile Thr Glu Lys Asp Ala Pro Gln Leu His Lys Leu |     |
| 85 90 95  |     |
| ctg aca aat atg att gag gat gcc ggc gat ctg gcc acc cgc agc gcg | 739 |
| Leu Thr Asn Met Ile Glu Asp Ala Gly Asp Leu Ala Thr Arg Ser Ala |     |
| 100 105 110   |     |
| aaa gag aaa tat atg cgc att cgc ccg ttt gcg ttc tac ggc gtt tca | 787 |
| Lys Glu Lys Tyr Met Arg Ile Arg Pro Phe Ala Phe Tyr Gly Val Ser |     |
| 115 120 125   |     |
| acc tgt aac act aaa gac cag gac aag ctg tcg aaa aac gga tct tac | 835 |
| Thr Cys Asn Thr Lys Asp Gln Asp Lys Leu Ser Lys Asn Gly Ser Tyr |     |
| 130 135 140   |     |
| cct tcc ggc cat acc tct acc ggt tgg gca acc gcg ctg gta ctg gcg | 883 |
| Pro Ser Gly His Thr Ser Thr Gly Trp Ala Thr Ala Leu Val Leu Ala |     |
| 145 150 155 160   |     |
| gag atc aat ccg cag cgg caa aac gaa att ctc aaa cgc ggc tat gaa | 931 |
| Glu Ile Asn Pro Gln Arg Gln Asn Glu Ile Leu Lys Arg Gly Tyr Glu |     |
| 165 170 175   |     |
| ttg ggc gaa agc cgg gtt atc tgc ggc tat cat tgg cag agc gat gtc | 979 |
| Leu Gly Glu Ser Arg Val Ile Cys Gly Tyr His Trp Gln Ser Asp Val |     |

|   | 180 | 185 | 190 |      |
|---|-----|-----|-----|------|
| gat gcg gcg cgg ata gtc ggc tcg gcg gtg gtg gcg acc ctg cat acc     |     |     |     | 1027 |
| Asp Ala Ala Arg Ile Val Gly Ser Ala Val Val Ala Thr Leu His Thr     |     |     |     |      |
|   | 195 | 200 | 205 |      |
| aac ccg gcc ttc caa cag cag ttg cag aaa gca aag gat gaa ttc gcc     |     |     |     | 1075 |
| Asn Pro Ala Phe Gln Gln Gln Leu Gln Lys Ala Lys Asp Glu Phe Ala     |     |     |     |      |
|   | 210 | 215 | 220 |      |
| aaa acg cag aag taacgtcatc gccgttgaac tcccggaggc ggcgcttaac         |     |     |     | 1127 |
| Lys Thr Gln Lys   |     |     |     |      |
| 225   |     |     |     |      |
| gcgccttctc cgggctacta aatcgcacag cgctgtagcc ccggtaagcg ccagcgccac   |     |     |     | 1187 |
| cggggatttt gagatagcca gcaccagtag tttcagccag cgtgatgaat acattaacgg   |     |     |     | 1247 |
| caggccgcat gagtcgtaga tactgttatac ggtttgcaac ttttttaagg ttttttcccg  |     |     |     | 1307 |
| gaggcggcgc gctgcgctt ctccgggcta ctaaatacgca cagcgctgta gccccggtaa   |     |     |     | 1367 |
| gcggcagcgc caccgggggt aacaagcgca gattcagaag cgcgtagcga acggcgcggt   |     |     |     | 1427 |
| atccgggdcgc gtaaacaatgg ttgatgcttt taactgcggc gtgccaaggt agaggaaacc |     |     |     | 1487 |
| gacaattttg tctgtttcgc ggcagccaaa gccttcgcgg acaaccggac tctcggttaa   |     |     |     | 1547 |
| cgcaccgata cgccagatac cgttatagcc ctgcgccact gcggccattt gcatcgccat   |     |     |     | 1607 |
| caccgcacat cccgcggaca tctcctgttc ccacagcggt acc                     |     |     |     | 1650 |

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| Met Lys Lys Arg Val Leu Ala Leu Cys Leu Ala Ser Leu Phe Ser Val |     |     |    |  |
| -20   | -15 | -10 | -5 |  |

|   |   |    |  |  |
|---|---|----|--|--|
| Asn Ala Phe Ala Leu Val Pro Ala Gly Asn Asp Ala Thr Thr Lys Pro |   |    |  |  |
| -1 1  | 5 | 10 |  |  |

|   |    |    |  |  |
|---|----|----|--|--|
| Asp Leu Tyr Tyr Leu Lys Asn Ala Gln Ala Ile Asp Ser Leu Ala Leu |    |    |  |  |
| 15  | 20 | 25 |  |  |



Leu Pro Pro Pro Pro Glu Val Gly Ser Ile Ala Phe Leu Asn Asp Gln  
 30 35 40

Ala Met Tyr Glu Lys Gly Arg Leu Leu Arg Asn Thr Glu Arg Gly Lys  
 45 50 55 60

Gln Ala Gln Ala Asp Ala Asp Leu Ala Ala Gly Asp Val Ala Asn Ala  
 65 70 75

Phe Ser Ser Ala Phe Gly Ser Pro Ile Thr Glu Lys Asp Ala Pro Gln  
 80 85 90

Leu His Lys Leu Leu Thr Asn Met Ile Glu Asp Ala Gly Asp Leu Ala  
 95 100 105

Thr Arg Ser Ala Lys Glu Lys Tyr Met Arg Ile Arg Pro Phe Ala Phe  
 110 115 120

Tyr Gly Val Ser Thr Cys Asn Thr Lys Asp Gln Asp Lys Leu Ser Lys  
 125 130 135 140

Asn Gly Ser Tyr Pro Ser Gly His Thr Ser Thr Gly Trp Ala Thr Ala  
 145 150 155

Leu Val Leu Ala Glu Ile Asn Pro Gln Arg Gln Asn Glu Ile Leu Lys  
 160 165 170

Arg Gly Tyr Glu Leu Gly Glu Ser Arg Val Ile Cys Gly Tyr His Trp  
 175 180 185

Gln Ser Asp Val Asp Ala Ala Arg Ile Val Gly Ser Ala Val Val Ala  
 190 195 200

Thr Leu His Thr Asn Pro Ala Phe Gln Gln Gln Leu Gln Lys Ala Lys  
 205 210 215 220

Asp Glu Phe Ala Lys Thr Gln Lys  
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<210> 13  
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<400> 13

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<400> 16

Asn Leu Ser Tyr Gly Asp Val  
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<400> 19

Asn Leu Ser Trp Gly Asp Val  
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<210> 21  
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<400> 22

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<400> 25

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<400> 28

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<400> 34

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<400> 35

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<400> 38

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25

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1 5

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25

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<400> 57  
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25

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mutation

<400> 58

Asn Leu Ser Gly Gly Asp Val  
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<400> 61

Asn Leu Ser His Gly Asp Val  
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Ala Asn Leu Trp Ser Gly Asp  
1 5

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<210> 69  
<211> 25  
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mutation

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Leu Ser Ser Trp Asp Val Ala  
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<220>  
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mutation

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Thr Asn Met Asp Glu Asp Ala  
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His Thr Ser Asn Gly Trp Ala  
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Gln Asp Lys Phe Ser Lys Asn  
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Gln Asp Lys Glu Ser Lys Asn  
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Leu Val Pro Ala Gly  
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Pro Ser Gly His  
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Ser Arg Xaa Xaa Xaa Xaa Xaa His Xaa Xaa Xaa Asp  
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<400> 124

Leu Ala Leu Val Ala Thr Gly Asn Asp Thr Thr Thr Lys Pro Asp Leu  
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Tyr Tyr Leu Lys Asn Ser Glu Ala Ile Asn Ser Leu Ala Leu Leu Pro

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|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Pro | Pro | Pro | Ala | Val | Gly | Ser | Ile | Ala | Phe | Leu | Asn | Asp | Gln | Ala | Met |
|     |     | 35  |     |     |     |     | 40  |     |     |     |     | 45  |     |     |     |
| Tyr | Glu | Gln | Gly | Arg | Leu | Leu | Arg | Asn | Thr | Glu | Arg | Gly | Lys | Leu | Ala |
|     | 50  |     |     |     |     | 55  |     |     |     |     | 60  |     |     |     |     |
| Ala | Glu | Asp | Ala | Asn | Leu | Ser | Ser | Gly | Gly | Val | Ala | Asn | Ala | Phe | Ser |
| 65  |     |     |     |     | 70  |     |     |     |     | 75  |     |     |     |     | 80  |
| Gly | Ala | Phe | Gly | Ser | Pro | Ile | Thr | Glu | Lys | Asp | Ala | Pro | Ala | Leu | His |
|     |     |     |     | 85  |     |     |     |     | 90  |     |     |     |     | 95  |     |
| Lys | Leu | Leu | Thr | Asn | Met | Ile | Glu | Asp | Ala | Gly | Asp | Leu | Ala | Thr | Arg |
|     |     |     | 100 |     |     |     |     | 105 |     |     |     |     | 110 |     |     |
| Ser | Ala | Lys | Asp | His | Tyr | Met | Arg | Ile | Arg | Pro | Phe | Ala | Phe | Tyr | Gly |
|     |     | 115 |     |     |     |     | 120 |     |     |     |     | 125 |     |     |     |
| Val | Ser | Thr | Cys | Asn | Thr | Thr | Glu | Gln | Asp | Lys | Leu | Ser | Lys | Asn | Gly |
|     | 130 |     |     |     |     | 135 |     |     |     |     | 140 |     |     |     |     |
| Ser | Tyr | Pro | Ser | Gly | His | Thr | Ser | Ile | Gly | Trp | Ala | Thr | Ala | Leu | Val |
| 145 |     |     |     |     | 150 |     |     |     |     | 155 |     |     |     |     | 160 |
| Leu | Ala | Glu | Ile | Asn | Pro | Gln | Arg | Gln | Asn | Glu | Ile | Leu | Lys | Arg | Gly |
|     |     |     |     | 165 |     |     |     |     | 170 |     |     |     |     | 175 |     |
| Tyr | Glu | Leu | Gly | Gln | Ser | Arg | Val | Ile | Cys | Gly | Tyr | His | Trp | Gln | Ser |
|     |     |     | 180 |     |     |     |     | 185 |     |     |     |     | 190 |     |     |
| Asp | Val | Asp | Ala | Ala | Arg | Val | Val | Gly | Ser | Ala | Val | Val | Ala | Thr | Leu |
|     |     | 195 |     |     |     |     | 200 |     |     |     |     | 205 |     |     |     |
| His | Thr | Asn | Pro | Ala | Phe | Gln | Gln | Gln | Leu | Gln | Lys | Ala | Lys | Ala | Glu |
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Phe Ala Gln His Gln Lys Lys  
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Leu Lys Asn Ala Gln Ala Ile Asp Ser Leu Ala Leu Leu Pro Pro Pro  
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Pro Glu Val Gly Ser Ile Ala Phe Leu Asn Asp Gln Ala Met Tyr Glu  
 35 40 45

Lys Gly Arg Leu Leu Arg Asn Thr Glu Arg Gly Lys Leu Ala Ala Glu  
 50 55 60

Asp Ala Asn Leu Ser Ala Gly Gly Val Ala Asn Ala Phe Ser Ser Ala  
 65 70 75 80

Phe Gly Ser Pro Ile Thr Glu Lys Asp Ala Pro Gln Leu His Lys Leu  
 85 90 95

Leu Thr Asn Met Ile Glu Asp Ala Gly Asp Leu Ala Thr Arg Ser Ala  
 100 105 110

Lys Glu Lys Tyr Met Arg Ile Arg Pro Phe Ala Phe Tyr Gly Val Ser  
 115 120 125

Thr Cys Asn Thr Thr Glu Gln Asp Lys Leu Ser Lys Asn Gly Ser Tyr  
 130 135 140

Pro Ser Gly His Thr Ser Ile Gly Trp Ala Thr Ala Leu Val Leu Ala  
 145 150 155 160



Glu Ile Asn Pro Gln Arg Gln Asn Glu Ile Leu Lys Arg Gly Tyr Glu  
165 170 175

Leu Gly Glu Ser Arg Val Ile Cys Gly Tyr His Trp Gln Ser Asp Val  
180 185 190

Asp Ala Ala Arg Ile Val Gly Ser Ala Val Val Ala Thr Leu His Thr  
195 200 205

Asn Pro Ala Phe Gln Gln Gln Leu Gln Lys Ala Lys Asp Glu Phe Ala  
210 215 220

Lys Thr Gln Lys  
225